REMARKS

Claims 1-20 were pending in the present application. Claims 10-20 were previously withdrawn from consideration as drawn to a non-elected invention. By virtue of this response, claims 1-2, and 5 have been amended and new claims 21-32 have been added. Accordingly, claims 1-9 and 21-32 are currently under consideration. Amendment and cancellation of certain claims is not to be construed as a dedication to the public of any subject matter of the claims as previously presented. Claim 2 has been amended to correct antecedent basis. The support for the amendment to claims 1 and 5 that recite "replication-defective" can be found at least at page 3, lines 24-26. Support for the amendment to claim 1 which recites adenoviral "sequence" can be found at least at page 5, line 1. Claim 5 has been amended for clarity. The support for new claim 21-23 can be found at least at the paragraph bridging pages 6-7. Support for claims 24-25 which recite human adenovirus can be found at least at the paragraph bridging pages 6-7. Support for new claims 26-27 can be found at least at page 6, lines 29-32. Support for new claim 28 can be found at least at page 11, lines 21-30. Support for new claims 29-32 can be found at least at page 11, lines 21-24.

Applicants request rejoinder of methods claims to the extent they incorporate all the limitations of allowed composition claims. In re Ochiai.

Attached hereto is a marked-up version of the changes made to the claims by the current amendment. The attached pages are captioned "**VERSION WITH MARKINGS TO SHOW CHANGES MADE**".

Concerning the objection of claims

Claim 6 has been objected to as being dependent upon a rejected base claim, but would be allowable if rewritten in independent form including all of the limitations of the base claim and any intervening claims.

Applicants acknowledge the Examiner's indication of allowable subject matter.

Concerning the rejection of claims under 35 U.S.C. § 112, second paragraph

Claim 2 stands rejected as allegedly indefinite for failing to particularly point out and distinctly claim the subject matter which applicants regard as the invention.

Applicants have amended claim 2 to delete reference to the adenosine A3 receptor, thereby obviating the Examiner's concerns.

Concerning the rejection of claims under 35 U.S.C. § 112, first paragraph

Claims 1-4 stand rejected because the specification, while being enabling for providing protection against simulated ischemia via adenovirus mediated gene transfer of a heat shock protein 70 (HSP70i) *in vitro*, wherein said adenovirus is a replication deficient adenovirus lacking E1A and E1B genes, allegedly does not reasonably provide enablement for providing protection against myocardial ischemia by using adenovirus mediated gene transfer of a heat shock protein via any administration route *in vitro* (Applicants believe the Examiner intended to recite *in vivo*), wherein said adenovirus is a replication deficient adenovirus lacking E1A and E1B genes. The Examiner alleges that the specification does not enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to use the invention commensurate in scope with these claims.

Applicants traverse this rejection of claims. To object to a specification on the grounds that the disclosure is not enabling, the Examiner must provide evidence or technical reasoning substantiating those doubts. The Examiner has not provided evidence or technical reasoning as to why the claimed invention is not enabled. Therefore, Applicants respectfully submit no *prima facie* case of non-enablement has been made.

Assuming *arguendo* that a *prima facie* case of non-enablement has been made, which Applicants don't concede, Applicants provide the following arguments. In order to satisfy the requirements of Section 112, first paragraph, a patent application must teach one of ordinary skill in the art how to make and use the claimed invention. It is well established that enablement is not precluded by the need for some experimentation. As stated in M.P.E.P. Section 2164.01, an analysis of whether a particular claim is supported by the disclosure in an application requires a

determination of whether that disclosure, when filed, contained sufficient information regarding the subject matter of the claims as to enable one skilled in the pertinent art to make and use the claimed invention, and whether the experimentation needed to practice the invention is undue or unreasonable. Applicants submit that it would not require undue or unreasonable experimentation for one of skill in the art to practice the claimed invention.

The Examiner alleges at page 4 of the Office Action that the claims read on gene therapy *in vivo* by using the claimed recombinant adenoviral vector in light of the specification. The specification teaches that the methods using the claimed adenoviral vectors provide myocardial protection in which the stress factor is produced and present in the myocardium at the time of the ischemia so as to protect the myocardium against subsequent ischemic episodes. See the specification at the paragraph at page 3, lines 17-32 through page 4, lines 1-13. The methods of the present invention using the claimed adenoviral vectors provide for expression of the replication-deficient adenoviral vector encoding the stress factor in cardiac myocytes, which do not undergo rapid turnover, at the time of ischemic episode. The replication-defective adenoviral vector encoding the heat shock protein is delivered directly to the myocardium of a patient, or for example, to donor hearts prior to cardiac transplantation. See the specification at page 14, lines 6-14. The replication-deficient adenovirus may be delivered prior to an anticipated event. See the specification page 4, lines 14-29. The replication-deficient vector can be delivered a plurality of times. See the specification at page 4, line 29.

The Examiner alleges that the specification fails to provide adequate guidance and evidence for delivering a recombinant adenoviral vector expressing a heat shock protein via different administration routes, such as oral administration, intravenous injection, intramuscular injection etc., to a particular myocardial site in a subject so as to provide protection against myocardial ischemia in a subject *in vivo*.

Applicants disagree with the Examiner's allegation. First of all, Section 112, first paragraph does not require evidence of clinical efficacy and does not require that the specification provide guidance as to specific administration routes, such as oral administration,

intravenous injection or intramuscular injection, in order for a claim to be enabled. Section 112, first paragraph requires that a patent application teach one of ordinary skill in the art how to make and use the claimed invention. The specification provides guidance as to the preparation of the claimed adenoviral vectors beginning at page 6, line 19 through page 9, line 20. The specification provides guidance regarding the use promoters, such as the specific ventricular myocyte specific promoter, to target myocytes. See the specification at page 9, lines 23-31. The specification also discloses the use of additional promoters at page 10, lines 1-11 and the CMV promoter at page 16, lines 18. The specification provides guidance as to administration of the claimed adenoviral vectors comprising a transgene encoding a heat shock protein. The specification teaches at page 11, lines 20-32 that the claimed adenoviral vectors can be in the form of an injectable preparation containing a pharmaceutically acceptable excipient. The specification provides the range of viral particles (10^{10}-10^{13}) which allows for effective gene transfer. The specification at page 3, lines 24-32; page 10, lines 26-28; and at the paragraph bridging pages 11-12 teach that the claimed adenoviral vectors can be delivered to a myocardium by direct intracoronary injection using catheters. For gene delivery to donor hearts prior to cardiac transplantation, the specification teaches the use of intracoronary injection. See the specification at page 14, lines 13-14. Therefore, the specification provides guidance as to making and using the claimed invention.

As basis for the Section 112, first paragraph rejection, the Examiner alleges that gene therapy was unpredictable at the time of the invention and that targeting to desired tissues *in vivo* continues to be unpredictable. As support for this Section 112, first paragraph rejection of claims, the Examiner relies on Deonarain (1998, Expert. Opin. Ther. Pat. vol. 8 pages 53-69) and Crystal (1995, Science, 270:404-410), as indicating that the problems hampering gene therapy include the ability to target a gene to a population of cells, express it at adequate levels for long enough periods of time, and regulate it. The Examiner also relies on Eck et al. (1996, Goodman and Gilman's The Pharmacological Basis of Therapeutics).

The references cited by the Examiner fail to provide specific evidence that one of ordinary skill in the art would not have been able to make and use the claimed invention without undue experimentation. The specification provides guidance as to preparing replication-defective adenoviral vectors; targeting adenoviral vectors to the myocardium, for example, by direct intracoronoary injection; administering the adenovirus and measuring protection against ischemic injury. The Examiner cites Deonarain and Crystal as describing "problems hampering gene therapy" such as targeting the gene to the desired population of cells and expressing the introduced gene for long enough periods of times. The specification provides guidance as to targeting the claimed adenoviral vectors to the myocardium and teaches that one of the advantages of using the claimed adenoviral vectors for transfer of heat shock proteins into the myocardium is the expression of the heat shock protein at the time of the ischemic event. See the specification at page 9, lines 3-9. The specification teaches at page 7, lines 8-9 that although the transgene is not passed onto daughter cells, this is not an important limitation and at lines 17-18 that transient gene transfer for some cardiovascular disease processes may be adequate and possible preferable.

Eck et al. define the goal of gene therapy at page 78 as being to genetically correct a defect in a part of the body. The specification teaches that the presently claimed adenoviral vectors are intended for administration to the myocardium with the goal of expression of the heat shock protein during the ischemic episode and encompasses the administration of the claimed adenoviral vectors to donor hearts, prior to cardiac transplantation. The invention is not directed to correcting a gene defect in the myocardium. Therefore, the Examiner's reliance on Deonarain, Crystal and Eck et al. in applying this Section 112, first paragraph rejection of claims appears misplaced.

The Examiner states that the specification does not provide any *in vivo* working examples. Applicants disagree. First, Section 112, first paragraph does not require any working examples to be present in the specification. Second, the specification provides enabling disclosure for the claimed invention. Experiment 3 provides an *in vivo* model to illustrate how

an adenoviral vector preparation containing the desired transgene can be injected into the heart of a large mammal (the pig, which is considered to be predictive of cardiac function in the human heart) and effectively target expression of the transgene in myocytes within the heart. Although the transgene used in Experiment 3 was β-galactosidase, the results demonstrated the effectiveness of the adenoviral vectors and delivery techniques in transferring genes to the myocardium of the large mammal heart. The experiment also demonstrated the lack of inflammation and other adverse effects on animals undergone the *in vivo* gene transfer procedure. See the specification at page 18, lines 5-28. There is no reason to doubt that another gene (such as the genes claimed) would not be expressed.

Experiment 4 further describes a pig constriction model to demonstrate *in vivo* adenoviral gene delivery of a stress factor transgene (exemplified by a heat shock protein HSP70i), and to assess the myocardial protective effects of the gene delivery. The materials and techniques used for the *in vivo* experiments have been described in previously published research papers. More importantly, Experiment 4 describes how to assess the myocardial protection effects of stress factor transgene expression.

The teaching of Experiment 4 is correlated by Experiment 5, which describes transfecting cardiomyocytes and myogenic cells *in vitro* with adenoviral vectors comprising a stress factor transgene (exemplified by a heat shock protein HSP70i). Experiment 5 demonstrates that (i) the target cells can be effectively transfected with the stress factor vectors; (ii) the stress factor transgene can be successfully expressed at an elevated level; and (iii) elevated expression of the stress factor transgene can mediate alterations at the cellular level which tend to protect such cells from the metabolic deprivations associated with cardiac ischemia. In view of these results obtained with the ischemic cell model using cardiac myocytes, one would not predict that expression of the stress factor transgenes in such cells *in vivo* would be incapable of providing a similar protective effect.

Additionally, Applicants submit the Declaration of Dr. Wolfgang Dillmann ("Dillmann Declaration") pursuant to 37 C.F.R. Section 1.132 which demonstrates that in an art accepted

animal model (which is considered predictive of cardiac function in the human heart), administration of heat shock protein (exemplified by heat shock protein 70i, HSP70i) from a replication-defective adenoviral vector administered to mouse myocardium *in vivo* protected against ischemic injury as measured by improvement in contractile performance and a decrease in creatine kinase, an indicator of ischemic injury after myocardial infarct in humans.

As disclosed in paragraphs 5-6 of the Dillmann Declaration, normal mice were anesthetized, placed on a ventilator and the heart was exposed and prepared for administration of the adenoviral vector. Aliquots of the replication-defective adenoviral vector containing nucleic acid encoding HSP70i under the control of CMV promoter, or adenoviral vector control (lacking nucleic acid encoding HSP70i operably linked to the CMV promoter) were injected into the left ventricular free wall. The hearts were repositioned back in the chest cavity, the chest cavity was sutured closed and the animals were allowed to recover for 4 days.

As disclosed in paragraph 7 of the Dillmann Declaration, at the end of 4 days, the animals were sacrificed, the hearts were excised and mounted on a Langendorff perfusion apparatus, and the hearts were subjected to a perfusion protocol comprising 15 minutes of aerobic perfusion followed by 20 minutes of global no-flow ischemia, and followed by 120 minutes of reperfusion.

As disclosed in paragraph 8 of the Dillmann Declaration, in mice injected with the adenoviral vectors expressing HSP70i, there was significant improvement in contractile performance as measured by maximally developed systolic pressure, the decrease in diastolic pressure and developed pressure. In additional experiments in the same animal model, it was demonstrated that the release of creatine kinase, an indicator of ischemic injury, was significantly decreased in the mouse hearts injected with the adenoviral vector expressing HSP70i.

The specification provides guidance as to how make and use the claimed invention. Additionally, the Dillmann Declaration provides experimental data demonstrating that in an art accepted animal model, administration of heat shock protein 70i (HSP70i) from a replication-defective adenoviral vector administered to mouse myocardium *in vivo* protected against

ischemic injury as measured by improvement in contractile performance and a decrease in creatine kinase. Therefore, in view of the arguments and evidence presented above, Applicants submit that the claimed invention is in full compliance with Section 112, first paragraph and respectfully request withdrawal of the Section 112, first paragraph rejection of claims.

Concerning the rejection of claims under 35 U.S.C § 103(a)

A. Claims 1-4 stand rejected under 35 U.S.C. § 103(a) as allegedly being unpatentable over Hayden et al., 1997 (US Patent No. 5,658,729) in view of Hutter et al., 1994 (Circulation, Vol.89, No. 1, p. 355-360) and Stege et al., (Experimental Cell Research, Vol. 214, No. 1, p. 279-284).

Applicants traverse this rejection of claims. Applicants do not agree or concede that a *prima facie* case of obviousness has been established and submit that the invention is non-obvious in view of the cited references. In order to establish a *prima facie* case of obviousness, there has to be, *inter alia*, some motivation or suggestion provided by the references, or in combination with the knowledge available to the skilled artisan, to modify the art cited or to combine reference teachings. Applicants submit that the combination of references cited does not provide motivation for arriving at the claimed invention, and, even if combined, the combination of references does not produce the claimed invention.

The presently claimed invention recites a replication-deficient recombinant adenoviral vector comprising a stress related factor which is a heat shock protein and methods of producing a replication-deficient adenoviral vector. Hayden et al. relates, in part, to identification of a single point mutation in the human lipoprotein lipase gene (LPL) that is seen with increased frequency in patients with coronary artery disease and, in part, to gene therapy to introduce functional LPL into a patient subject to a defect in LPL. As concluded by the Examiner, there is no teaching or suggestion whatsoever in Hayden et al. of a recombinant, replication-deficient adenoviral vector expressing a heat shock protein. Furthermore, there is no teaching, suggestion or appreciation in Hayen et al., of the use of a replication-deficient adenoviral vector expressing a heat shock protein in protecting the myocardium from ischemic events. Hutter et al. relate to *in vivo* heat-shock protein induction in rat hearts pre-treated with whole body hyperthermia. Hutter et al. have no teachings or suggestions whatsoever regarding any form of an adenovirus vector, much less, the administration of a replication-defective adenoviral vector comprising a transgene

coding for heat shock protein to the myocardium in order to provide protection from ischemic events. The Examiner alleges that Stege et al. teach the presence of human HSP72 and the use of HSP72 in transfecting Rat-1 fibroblast cells. Stege et al. have no suggestion whatsoever regarding replication-deficient adenovirus vector comprising nucleic acid encoding a heat shock protein. Furthermore, there is no suggestion whatsoever in any of the references to combine them. There is no suggestion in Hayden et al., which relates to a single point mutation in LPL, to combine with the teachings of Hutter et al., which relate to rat heart, heat shock protein induction or Stege et al., which disclose Rat-1 fibroblast cells transfected with HSP72. That the Examiner would combine these references to allege non-obviousness is the result of the impermissible use of hindsight reconstruction. Even if the references are combined, one of skill in the art would not arrive at the presently claimed invention.

The presently claimed invention is non-obvious in view of the art cited by the Examiner and Applicants respectfully request withdrawal of the Section 103(a) rejection of claims.

B. Claims 5 and 7-9 stand rejected under 35 U.S.C. § 103(a) as allegedly being unpatentable over Hayden et al., 1997, Hutter et al., 1994, and Stege et al., 1994 as applied to claims 1-4 above, and further in view of McGrory et al., 1988 (Virology, Vol. 163, p. 614-617).

Applicants traverse this rejection of claims. The presently claimed invention is not rendered obvious by the references cited by the Examiner.

Claims 5 and 7-9 recite methods of producing a replication-deficient adenoviral vector. As discussed above, none of Hayden et al., Hutter et al., or Stege et al. have any suggestions whatsoever regarding the claimed invention, that is, a replication-deficient adenoviral vector comprising a heat shock protein and methods of making such a vector. McGrory et al. relates to rescue of early region 1 mutations into infectious human adenovirus. McGrory et al. have no suggestions whatsoever regarding a replication-deficient adenoviral vector expressing a heat shock protein much less of the use of such a vector in protecting the myocardium from ischemic events. McGrory et al. in no way cures the deficiencies of Hayden et al., Hutter et al., or Stege et al. Furthermore, there is no suggestion in any of the references to support the combination and if combined yield no motivation and no expectation of successfully producing the claimed

invention. The presently claimed invention is not rendered obvious by the references cited by the Examiner. In view of the arguments above, Applicants respectfully request withdrawal of the Section 103(a) rejection of claims.

CONCLUSION

Applicants have made a sincere effort to overcome the rejections and address all issues that were raised in the outstanding Office Action. Accordingly, reconsideration and allowance of the pending claims are respectfully requested. If it is determined that a telephone conversation would expedite the prosecution of this application, the Examiner is invited to telephone the undersigned at the number given below.

In the unlikely event that the transmittal letter is separated from this document and the Patent Office determines that an extension and/or other relief is required, applicants petition for any required relief including extensions of time and authorizes the Assistant Commissioner to charge the cost of such petitions and/or other fees due in connection with the filing of this document to **Deposit Account No. 03-1952** referencing docket no. 220002057202.

Respectfully submitted,

Dated: July 13, 2002

By: _____

Debra J. Glaister
Registration No. 33,888

Morrison & Foerster LLP
755 Page Mill Road
Palo Alto, California 94304-1018
Telephone: (650) 813-5725
Facsimile: (650) 494-0792

VERSION WITH MARKINGS TO SHOW CHANGES MADE

In the Claims:

Claims 1-2 and 5 have been amended, as follows:

1. (Twice amended) An isolated [and purified recombinant], replication-deficient adenoviral vector, said vector comprising:

an adenoviral sequence [genome] from which the E1A/E1B genes have been deleted;

a transgene coding for a stress related factor which is a heat shock protein; and

a promoter operably linked to said transgene, wherein expression of the transgene is controlled by said promoter.

2. (Once amended) The vector of claim 1, wherein said stress related factor is selected from the group consisting of HSP70i, HSP27, HSP40, and HSP60[, and the adenosine A3 receptor].

5. (Once amended) A method of producing [an isolated and purified] a recombinant replication-deficient adenoviral vector comprising a transgene coding for a stress related factor [vector of claim 1], comprising the steps of:

[cloning a transgene coding for a stress related factor into a plasmid containing a promoter and a polylinker flanked by adenoviral sequences of the left end of the human adenovirus 5 genome from which the E1A/E1B genes have been deleted;]

co-transfecting [said] a plasmid comprising a transgene coding for a stress related factor, a promoter and a polylinker flanked by adenoviral sequences of the left end of the human adenovirus 5 genome from which the E1A/E1B genes have been deleted into a mammalian cell[s] transformed with [the]E1A/E1B genes, with a plasmid which contains the entire human adenoviral 5 genome, and an additional insert making the plasmid too large to be encapsulated, whereby rescue recombination takes place between the transgene-inserted plasmid and the plasmid having the entire adenoviral genome so as to create a recombinant genome containing the transgene without the E1A/ElB genes, said recombinant genome being sufficiently small to be encapsulated;

identifying cells comprising recombinant vectors in cell cultures;

propagating the resulting recombinant vectors in mammalian cells transformed with the E1A/E1B genes; and

purifying the propagated recombinant vectors.

New claims 21-32 have been added.